Efficacy of Bioagents against *Alternaria porri* Incitant of Purple Blotch of Onion in Egypt M.F.A. Ahmed*, M.M. Amin** and I.A.I. El-Fiki***

*Central Lab. Organic Agric., A.R.C., Egypt. **Plant Pathol. Res. Inst., ARC, Giza, Egypt. ***Plant Pathol. Dept., Fac. Agric., Benha Univ., Egypt.

E fficacy of *Trichoderma harzianum*, *T. hamatum*, *T. viride*, and *Chaetomium* sp. as foliar spray against purple blotch of onion (*Allium cepa*) was evaluated. The fungicides Blight stop (bio) and Luna Experience SC (chemical) at certain doses were used as a check. All treatments recorded the highest efficacy percentage for reducing severity of purple blotch compared to control, where it gave 88.85 and 69.76% after four and seven sprays respectively. *Trichoderma harzianum* was the superior treatment more than Blight stop as a check. On the other hand, *Chaetomium* sp. showed the lowest effect. Also, all treatments increased onion yield, plant height, number of leaves and fresh weight/plant, total carbohydrates, total nitrogen and total soluble solids as compared to non-treated plants.

Keywords: Alternaria porri, Allium cepa, biological control, Chaetomium sp., onion, purple blotch and Trichoderma spp.

Onion (*Allium cepa* L.) is one of the oldest known and important crops grown in Egypt and many countries of the world. It has been reported to be rich in phytochemicals especially medicinal flavonoids (Javadzadeh *et al.*, 2009). Purple blotch caused by *Alternaria porri* is the most prevalent and dangerous foliar disease worldwide and causes the major problems of onion production in Egypt (Abdel-Megid *et al.*, 2001). Yield losses reached sometimes to 97.3% (Lakra, 1999). These losses mainly result from severe infections in bulb onion crops causing early defoliation, reduced bulb size, and poor storage quality of bulbs (Surviliene *et al.*, 2008). Purple blotch disease is one of the most destructive diseases of *Allium species* in Egypt. The disease is more severe for seed crops in comparison to bulb crops, sometimes causing 100% loss of onion seed production (Abdel-Rahim *et al.*, 2016, Gupta and Pathak, 1988 and Quadri *et al.*, 1982). The high relative humidity up to 100% and temperature range from 20-28 °C were optimum for purple blotch infection (Kumar, 2007), causing sometimes 100% loses of the seed production (Daljeet *et al.*, 1992 and Schwartz, 2004).

Chemical control of onion purple blotch had been practiced by many researchers (Rahman *et al.*, 1988 and Hasan *et al.*, 2016). The harmful side effects of fungicides such as carcinogenic residual, toxicity problems, environmental pollution and development of fungicide-resistant strains of pathogens were reported (Rial-Otero *et al.*, 2005). Additionally, use of synthetic fungicides can cause development of

fungicide-resistant strains as well as the appearance of diseases which their severity is increased by the use of specific chemical product has stimulated. Therefore, a large number of studies have been devoted to searching for alternative control strategies to plant pathogens (Vijayalakshmi *et al.*, 2012).

Biological control through the use of antagonistic microorganisms is a potential non chemical means of controlling plant disease by reducing inoculum levels of pathogens. Such management would help prevention of the pollution and also health hazards (Kumar, 2007). Several researchers have reported biological control and effective antagonistic potential of both fungal and bacterial antagonistic microorganisms for controlling onion purple blotch (Chincholkal and Mukerji, 2007, Kumar, 2007 and Fayzalla *et al.*, 2011). Trichoderma isolates showed mycoparasitic activity and competitive capability against the mycelial growth of *A. porri* and caused 79.5% disease reduction (Chethana *et al.*, 2012). *T. harzianum* was found to be effective and recorded least mean disease intensity (25.54%) of *A. porri* and highest onion bulb yield (33.01 t/ha) in comparison with control treatment (Chethana *et al.*, 2015).

The present work aimed to control purple blotch disease of onion through finding out the most suitable bioagent treatment that has the ability to protect onion plants against purple blotch disease under field conditions and maintain high quality and quantity of onion crop.

Materials and Methods

Laboratory studies:

a. Isolation and purification of causal organism of purple blotch of onion:

Diseased samples of naturally infected leaves of onion (*Allium cepa* L.) were collected from growing areas of Bio Land Farm, Sharkiya governorate, Egypt. The infected leaves of each sample were thoroughly washed with tap water, air dried, cut into small pieces, each piece contains single lesion, surface disinfected with Sodium hypochlorite (5 % active chlorine) for 2 minutes, washed several times with sterilized distilled water and dried between two sterilized filter papers. Small portions of spotted lesions were placed in Petri dishes containing Gliotoxin fermentation agar (GFA) medium (Brian and Hemming, 1945) at the rate of five pieces/plate and incubated at $25\pm2^{\circ}$ C for 4-7 days. The growing fungi were examined microscopically and purified using hyphal tips or single spore method (Riker and Riker, 1936). Either hyphal tips or single spores were carefully transferred to slopes of GFA medium. Pure cultures of each isolate were maintained on GFA slant and kept at 4°C for identification and further experiments.

b. Identification of the causal organisms:

Identification of the isolated fungi was carried out at the Plant Pathology Dept., Fac. Agric., Moshtohor, Benha Univ. according to their cultural, morphological and microscopic characteristics as described by Singh (1982) and Barnett and Hunter (1987).

c. Antagonistic microorganisms:

Chaetomium sp. was obtained from Onion, Garlic and Oil Crops Dept., Plant Pathol. Res. Inst., ARC. While, *Trichoderma harzianum*, *T. hamatum*, *T. viride* were kindly obtained from Central Lab. of Organic Agriculture, CLOA, ARC.

Blight stop

The recommended bio fungicide consists of *Trichoderma* spp. 30×10^6 was obtained from CLOA and used in this investigation as a comparison with the tested treatments.

Luna Experience SC

The recommended fungicide Luna Experience SC (Luna) was used in this investigation as a comparison with the tested treatments. The fungicide is manufactured by Bayer, Crop Science, Germany, consists of Fluopyram 17.6% and Tebuconazole 17.6%.

d. Antagonistic potentiality of the antagonistic fungi against Alternaria porri

Petri dishes 9cm, each contains 10 ml of Gliotoxin Fermentation Agar (GFA) medium were inoculated at one side with 5mm agar disc obtained from the periphery of *Alternaria porri* colonies (5-days old), grown on the same medium. Whereas, the other side of the plate was inoculated with a similar agar disc obtained from the antagonistic fungal culture, *i.e. Trichoderma harzianum*, *T. hamatum*, *T. viride* and *Chaetomium* sp. Three plates were used for each particular treatment. Plates inoculated only at one side of petri dishes, with a known pathogenic fungus were used as a control treatment. Inoculated plates were incubated at $25\pm2^{\circ}$ C. The experiment was terminated when mycelial growth covers the medium surface in any plate of the treatments. All plates were examined and the percentage of reduction in mycelial growth of the pathogenic fungi was determined using the formula suggested by Ahmed (2013):

% Reduction in linear growth = $100 - [(G2 / G1) \times 100]$

Where: G1: growth of the pathogenic fungi in plates inoculated with the pathogen alone.

G2: growth of the pathogen against the antagonist.

e. Preparation of the biocontrol agents

In this experiment, suspensions containing propagules of the tested biocontrol agents, *i.e. Chaetomium* sp., *Trichoderma harzianum, T. hamatum, T. viride* were prepared as follows: Each of the antagonistic fungi was grown for 10 days at $25\pm2^{\circ}$ C in liquid Gliotoxin fermentation (GF) medium under complete darkness conditions (Ahmed, 2013). All cultures were individually blended in an electrical blender for 2 minutes then, used as suspension at concentration of 30×10^{6} spores/ml with dilution 1:100.

Field application of different treatments

Field experiments were carried out at the Bio Land Farm, Sharkiya governorate, Egypt during 2014/2015 and 2015/2016 winter growing seasons. Experiments were designed as randomized complete blocks. Three plots were used as replicates for all treatments and control. The area for each plot was $10.5m^2$ (3.0 X 3.5m). Onion

transplants sixty days old, Giza 20 and Giza Red were planted on 1^{st} of December. The recommended agricultural practices and irrigation for onion crop were used. *Chaetomium* sp., *T. harzianum*, *T. hamatum*, *T. viride* and Blight stop were used as suspensions at concentration of 30×10^6 spore/ml with dilution 1:100, while Luna fungicide was used at the rate of 1ml/L. Arabic gum and potassium soap were added at 5% for each spray.

Only distilled water was applied in the same period to act as control. Foliar spraying the treatments was applied at regular interval periods and repeated 7 times every two weeks (duration from January, 15 to April, 15). All treatments received the same normal agricultural practice till harvest at May.

Determination of biochemical components:

Some bio constituents were carried out on the two investigated onion verities at the end of each season. Total carbohydrates were determined in the aqueous extract according to Dubois *et al.* (1956). Total nitrogen percentage was determined according to Jackson (1967). Total soluble solids (TSS) were determined using a Carlzeiss hand Refractometer.

Disease assessments:

One hundred leaves from each plot were randomly collected to determine disease severity after 3 and 7 sprays and were monitored using (0-5) scale and recorded according to the method described by Sharma (1986) as follows:

0 = no infection (leaves are completely healthy), 1 = a few spots towards the tip covering less than 10% of leaf area, 2 = several dark purplish brown patches covering up to 20% of leaf area, 3 = several patches with paler outer zone covering up to 40% of leaf area, 4 = leaf streaks covering up to 75% of leaf area or breaking of the leaves from the center, and 5 = complete drying of the leaves or breaking of the leaves from the base. Disease severity index of purple blotch was estimated using the following formula:

D.S.I%=
$$\frac{\Sigma (n xv)}{ZN} \times 100$$

Where:

D.S.I= Disease severity index, n = Number of leaves in each category, v = Numerical value of each category, z = Numerical value of highest category and N = Total number of leaves in the sample.

In addition, plant height (cm), No. of leaves/plant, fresh weight/plant (g) were determined and onion bulb yield as kg/plot was weighted at the end of each season.

Statistical analysis

Data were subjected to statistical analysis and compared according to the least significant difference (LSD) as mentioned by Snedecor and Cochran (1989).

Results and Discussion

Isolation and identification of the causal organism:

The infected leaves were observed for purple blotch symptoms. On infected leaves, small, deep, oval to foot-ball shaped lesions were found. The lesions are brown to purple at the centre surrounded by a light brown area. The pathogen was isolated from the infected leaves of onion and cultured on GFA agar medium. The young hyphae are hyaline, slender, radiating and septate. The conidiophores arose singly or in groups, pale brown, erect, simple, cylindrical and septate. Conidia are solitary, straight or curved with the body of conidium ellipsoidal tapering to the beak and having 7 to 9 transverse septa and 1 to 3 longitudinal septa. White colonies turned purple color with advancing age of culture. With the above characteristics, the pathogen was identified as *Alternaria porri* according to the description of Singh (1982) and Barnett and Hunter (1987).

Antagonistic potentiality of the antagonistic fungi against Alternaria porri:

This experiment was carried out to evaluate the effect of different antagonistic isolates against the mycelial growth of *Alternaria porri*. Data in Table 1 indicate that the different antagonistic isolates varied in their effect against the tested fungus. All tested antagonists caused significant reduction in the linear growth of *A. porri*. Data in the same Table 1 show that *T. harzianum* caused the highest reduction (85.2 %, on the average) followed by *T. viride* (82.8 %), *T. hamatum* (78.4 %) and *Chaetomium* sp. (71.8 %). These results are in harmony with those previously recorded by Elad (2000) and Ahmed (2013). The pronounced antifungal activity of *Trichoderma* spp., may be attributed to some lytic enzymes, which act as fungal cell-wall-degrading agents such as N-acetyl- β -D-glucosedeaminidase, chitinase, β -1,3 gluconase, chitobiosidase and protease (Abo-Elyousr *et al.*, 2014 and Ahmed *et al.*, 2016).

uays	
Antagonists	% Reduction in growth of Alternaria porri
Chaetomium sp.	71.80
T. harzianum	85.20
T. hamatum	78.40
T. viride	82.80
Control "Untreated"	00.00
L.S.D at 1%	1.32

Table 1. Effect of the antagonistic fungi on the percentage of reduction in linear growth of *Alternaria porri* after incubation at 25±2°C for 5 days

Disease severity:

Data in Table 2 indicate that all tested bio-agents (*Chaetomium* sp., *Trichoderma* harzianum, T. viride and T. hamatum) significantly reduced disease severity of purple blotch on Giza 20 and red cultivars in 2014/2015 and 2015/2016 growing seasons compared to non-treated plants. In this regard, the different antagonistic isolates varied in their effect against disease parameters. According to the mean value of two seasons T. harzianum was the best treatment in disease reduction during the two successive seasons. Trichoderma harzianum had the superiority of Blight stop and showed no significant differences with Luna experience. While, *Chaetomium* sp. showed the lowest efficacy in reducing the disease severity during both growing seasons. On the other hand, Giza red cultivar was more sensitive than Giza 20 during the two seasons. Cultivars of onion differed significantly in their reaction to purple blotch and could therefore, be incorporated in breeding for disease resistance.

The results may be explained according to the dual effect of bio-agents which produce growth regulators in addition to the chemical effect of antioxidant, which play a clear role in improving plant physiology, metabolism (Harman, 2006) and induced systemic resistance (Harman *et al.*, 2004). Different mechanisms have been suggested as being responsible for their biocontrol activities, which can be divided into direct and indirect effects on the plant pathogen.

under field conditions during 2014/15 and 2015/10 growing seasons.									
		% Efficacy of treatments on disease severity of purple blotch							
Cultivar 7	Treatment	After three sprays				After seven sprays			
		2014/15	2015/16	Mean	Efficacy**	2014/15	2015/16	Mean	%Efficacy**
	Chaetomium sp.	4.6	5.2	4.9	65.8	9.9	13.3	11.6	38.5
	T. harzianum	1.5	1.7	1.6	88.8	4.1	7.3	5.7	69.8
	T. hamatum	3.1	3.5	3.3	77.0	9.1	11.6	10.3	45.1
Giza 20	T. viride	2.5	2.7	2.6	81.9	7.3	9.1	8.2	56.5
	Blight stop	2.2	2.5	2.3	83.6	6.8	8.5	7.6	59.4
	Luna	1.3	1.4	1.3	90.6	3.8	5.0	4.4	76.7
	Control	11.6	17.1	14.3	-	15.6	22.1	18.8	-
LSD at 5%		0.5	0.5	-	-	2.1	0.6	-	-
Giza red	Chaetomium sp.	5.1	5.8	5.4	73.9	12.3	15.7	14.0	47.8
	T. harzianum	2.3	2.8	2.5	87.8	5.8	6.5	6.1	77.0
	T. hamatum	4.4	4.6	4.5	78.4	9.8	10.3	10.0	62.5
	T. viride	3.4	3.5	3.4	83.4	7.1	9.1	8.1	69.8
	Blight stop	2.7	3.2	2.9	85.8	6.2	7.3	6.7	74.8
	Luna	1.9	2.1	2.0	90.4	4.7	5.8	5.2	80.4
	Control	19.4	22.3	20.8	-	23.4	30.2	26.8	-
LSD at 5%		0.72	0.96	-	-	3.9	1.3	-	-
* 0									

Table 2. Effect of different bio-treatments and fungicides as foliar spray [*] on
disease severity of purple blotch on Giza20 and Giza red onion plants
under field conditions during 2014/15 and 2015/16 growing seasons.

* Spray every two weeks beginning from January, 15 to April, 15 (Seven sprays)

** % Efficacy = ((control-treatment)/control)×100

Direct effects include suppression, competition for nutrients or space, production of antibiotic and lytic enzymes, inactivation of the pathogen's enzymes, parasitism, hypovirulence and predation. Indirect effects include all those aspects that produce morphological and biochemical changes in the host plant to induce resistance (Harman, 2006; Abo-Elyousr *et al.*, 2014 and Silva *et al.*, 2001).

Effect of different antagonists on some vegetative growth characteristics:

Concerning the influence of disease control treatments on vegetative growth, data in Table 3 show that all the studied vegetative growth parameters, *i.e.*, plant height, number of leaves and fresh weight/plant of the two cultivars were significantly increased compared to non-treated plants. *Trichoderma harzianum*, during both growing seasons was more effective than Blight stop and in most cases showed no differences with Luna fungicide.

These results are in agreement with those reported by Harman (2006) and Abo-Elyousr *et al.* (2014). Results of the present study showed that *Trichoderma* spp. improved the aforementioned crop parameters during the two growing seasons. Growth stimulation may be due to regulators such as IAA (Prakasam and Sharma, 2012).

50	easons								
	Treatment	Vegetative growth characteristics of onion plants							
		2014/15	growing s	eason	2015/16 growing season				
Cultivar		Plant height (cm)	No. of	Fresh	Plant	No. of	Fresh		
			leaves/	weight/	height	leaves	weight /		
		(em)	plant	plant (g)	(cm)	/plant	plant (g)		
	Chaetomium sp.	75.00	7.4	220.33	78.33	7.2	219.00		
	T. harzianum	90.60	10.0	276.67	87.66	9.6	245.66		
	T. hamatum	81.00	8.8	223.10	80.00	8.5	224.50		
Giza 20	T. viride	85.67	9.4	235.00	81.33	9.0	229.00		
	Blight stop	88.33	9.6	256.33	85.67	9.4	242.67		
	Luna	98.00	12.4	286.67	88.33	11.8	254.67		
	Control	64.00	6.5	154.00	60.00	6.3	122.50		
L.S.D. at 5	L.S.D. at 5%		0.87	15.83	7.44	0.75	19.79		
	Chaetomium sp.	71.00	6.4	213.17	70.00	6.0	194.00		
	T. harzianum	87.00	7.6	236.67	81.33	7.4	228.00		
Giza red	T. hamatum	79.30	6.8	220.83	72.33	6.5	198.38		
	T. viride	81.00	7.0	232.50	76.66	6.8	203.13		
	Blight stop	84.33	7.2	234.50	79.67	7.0	210.00		
	Luna	88.33	8.0	236.67	84.33	7.8	229.67		
	Control	60.00	6.0	146.00	56.00	5.8	157.00		
LSD at 5%	LSD at 5%		2.08	23.77	7.71	1.72	19.93		

Table 3: Effect of different bio-treatments and fungicides as foliar spray* on
some vegetative growth characteristics of Giza20 and Giza red onion
plants under field conditions during 2014/15 & 2015/16 growing
seasons

* Spray every two weeks beginning from January, 15 to April, 15 (Seven sprays)

The diversity of mechanism available to *Trichoderma* sp. for pathogen suppression through broad range of antifungal metabolites production, mycoparasitism, competition with pathogen of nutrient, occupation of infection court and induced resistance (Elad, 2000). Kumar and Palakshappa (2008) also reported significant increase in seed germination, vigour index and fresh weight of seedling over untreated control by seed treatment with *T. viride* against *A. alternata* on onion. *T. harzianum* Th-3 isolate effectively managed the pathogen and simultaneously increased the growth of onion plants.

Onion yield and bulb diameter:

Results shown in Table 4 indicate that applying any of the tested antagonists for treating onion as foliar application led to significant increase in both onion yield and bulb diameter for the two tested cultivars on 2014/2015 and 2015/2016 growing seasons compared to non-treated plants. *Trichoderma harzianum* caused the highest significant increase in the bulb diameter and total yield. Also, *T. harzianum* was superior on Blight stop and showed no significant differences with the fungicide Luna experience. On the other hand, *Chaetomium* sp. was the least effective one during the two growing seasons. Concerning onion cv. Giza 20, produced the highest value of onion bulb diameter and total bulb yield more than Giza red.

		Bulb diameter and yield components of onion					
Cultivars	Treatments	2014/15 growi	ng season	2015/16 growing season			
		Bulb diameter (mm)	Onion yield (kg/plot)	Bulb diameter (mm)	Onion yield (kg/plot)		
	Chaetomium sp.	63.0	36.4	61.4	33.9		
	T. harzianum	72.12	43.5	70.1	40.7		
	T. hamatum	66.4	37.6	62.4	36.7		
Giza 20	T. viride	68.9	39.6	67.5	38.2		
	Blight stop	71.22	42.0	70.0	39.7		
	Luna	75.25	44.2	72.2	42.0		
	Control	58.5	13.9	57.0	13.2		
L.S.D. at 5%	L.S.D. at 5%		5.2	3.3	2.9		
	Chaetomium sp.	62.9	34.5	61.0	33.5		
Giza red	T. harzianum	71.1	41.1	70.0	40.7		
	T. hamatum	64.1	37.2	61.9	35.8		
	T. viride	68.5	39.1	65.0	37.4		
	Blight stop	70.9	40.5	68.9	40.5		
	Luna	73.2	43.1	71.2	42.4		
	Control	57.4	13.2	56.9	11.5		
LSD at 5%		2.1	23.8	7.7	1.72		

 Table 4. Effect of different bio-treatments and fungicides as foliar spray^{*} on bulb diameter and yield components of Giza 20 and Giza red onion plants grown under field conditions during 2014/2015 and 2015/2016 growing seasons

* Spray every two weeks beginning from January, 15 to April, 15 (Seven sprays)

These results are in agreement with those reported by Kumar (2007), Fayzalla *et al.* (2011), Chethana *et al.* (2012), Chethana *et al.* (2015) and Ahmed *et al.* (2016). Also, Prakasam and Sharma (2012) reported that bulb diameter was increased and found anti-fungal compounds in onion bulb grown with Trichoderma treatment by chromatography. The efficiency of *T. harzianum* (Th-3) as a potential bio-control agent against, *A. porri* pathogen indicates the ability of protection and production of *T. harzianum* (Th-3) to serve as model for environment friendly biocontrol agent. Th-3 effectively managed the pathogen and simultaneously increased the growth of plants and improved yield of both onion bulb and seed crop (Kumar and Palakshappa, 2008).

Effect of different treatments on chemical components of onion:

Data concerning these analyses are presented in Table 5 beside those mentioned in Table 4 about the effect of these treatments on bulb diameter and yield just to correlate and explain the role of these treatments in changes that may be occurred in plant chemical components "total carbohydrates, total nitrogen (%) and TSS" and reflection of these changes on degree of resistance or increase in yield.

Trichoderma harzianum was the highest effective treatment followed by Blight stop as bio-treatment which led to the highest amount of plant chemical components compared with control treatment during the two successive growing seasons for the two cultivars under investigation.

-
Table 5. Effect of different bio-treatments and fungicides as foliar spray [*] on chemical
components of Giza 20 and Giza red onion plants grown under field conditions
during 2014/15 and 2015/16 growing seasons

during 2014/15 and 2015/10 growing seasons								
	Treatment	Chemical components of onion						
Cultivar		Total car	bohydrates	Total nitrogen		TSS		
		(mg /g dry weight)		(%)		(%)		
		2014/15	2015/16	2014/15	2015/16	2014/15	2015/16	
	Chaetomium sp.	1.45	1.223	12.67	12.33	12.57	12.07	
	T. harzianum	1.56	1.533	14.17	13.56	14.57	14.47	
	T. hamatum	1.46	1.347	12.90	12.52	12.93	12.83	
Giza 20	T. viride	1.46	1.377	13.15	12.71	13.70	13.50	
	Blight stop	1.48	1.407	13.37	13.21	14.53	14.07	
	Luna	1.86	1.553	16.08	15.94	15.20	15.07	
	Control	0.17	0.163	8.60	8.12	11.27	11.18	
L.S.D. at 5%		0.12	0.116	0.64	0.79	0.67	0.60	
Giza red	Chaetomium sp.	1.34	1.177	12.58	11.33	12.07	12.07	
	T. harzianum	1.53	1.490	13.56	13.00	14.43	14.13	
	T. hamatum	1.41	1.347	12.87	11.90	12.90	12.63	
	T. viride	1.46	1.373	13.06	12.65	13.30	13.13	
	Blight stop	1.52	1.427	13.35	12.75	13.90	13.53	
	Luna	1.82	1.533	15.94	15.40	15.17	14.67	
	Control	0.16	0.153	8.75	7.75	11.13	10.83	
LSD at 5%		0.12	0.104	0.60	0.73	0.66	0.52	

* Spray every two weeks beginning from January, 15 to April, 15 (Seven sprays)

On the contrary, *Chaetomium* sp. was the least effective treatment which led to the least content in plant chemical component rather than control treatment. The fungicide Luna Experience SC as check control, was more effective than all other treatments.

These results are in harmony with those obtained by Chethana *et al.* (2012), Chethana *et al.* (2015) and Ahmed *et al.* (2016). Treatments with high effect on plant protection and disease reduction were combined with increase in amount of total charbohydrates, total nitrogen (%) and TSS. These results are in agreement with those obtained by Kumar (2007) and Fayzalla *et al.* (2011).

References

- Abdel-Megid, M.S.; Metwally, A.H.; Abdel-Momen, S.M. and Hilal, A.A. 2001. A preliminary field study on the possibility of controlling foliar diseases of onion using some Egyptian medicinal plant extracts in comparison with a fungicide. *Egypt. J. Phytopathol.*, **29**(1): 21-31.
- Abdel-Rahim, I.R.; Abdel-Hafez, S.I.I. and Abo-Elyousr, K.A.M. 2016. Purple Blotch Disease of Onion Plant (*Allium cepa* L.) in Assiut, Egypt. LAP Lambert Academic Publishing, 192p.
- Abo-Elyousr, K.A.M.; Abdel-Hafez, S.I.I. and Abdel-Rahim, I.R. 2014. Isolation of *Trichoderma* and evaluation of their antagonistic potential against *Alternaria porri. J. Phytopathol.*, 162(9): 567-574.
- Ahmed, M.F.A. 2013. Studies on non-chemical methods to control some soil borne fungal diseases of bean plants *Phaseolus vulgaris* L. Ph.D. Thesis. Faculty of Agriculture, Cairo Univ., 137p.
- Ahmed, M.F.A.; Zayan, S.A.M. and Rashed, M.S. 2016. Evaluation of seed coating with some bio agents against damping-off and root rot diseases of fennel under organic farming system. J. Phytopathol. Pest Management, 3(3): 11-23.
- Barnett, H.J. and Hunter, B.B. 1987. Illustrated Genera of Imperfect Fungi. Burgess, Publ.Co., Minneapolis, USA, 218p.
- Brian, P.W. and Hemming, H.G. 1945. Gliotoxin a fungistatic metabolic product of *Trichoderma viride. Ann. Appl. Biol.*, 32:214-220.
- Chethana, B.S.; Ganeshan, G.; Archana, S.R. and Bellishree, K. 2015. Integrated disease management of purple blotch of onion. *Bioscan Int. Quartarly J. Life Sci.*, 10(4): 1729-1733.
- Chethana, B. S.; Ganeshan, G.; Rao, S.A. and Bellishree, K. 2012. In Vitro evaluation of plant extracts, bioagents and fungicides against Alternaria porri (Ellis) Cif., causing purple blotch disease of onion. Pest Management Hort. Ecosystems, 18:194-198.

- Chincholkal, S.B. and Mukerji, K.G. 2007. Biological Control of Plant Disease. Haworth Food and Agricultural Products Press TM, New York. London and Oxford: 223-228.
- Daljeet, S.; Dhiman, J.S.; Sidhu, A.S. and Singh, H. 1992. Current status of onions in India: strategies for disease resistance breeding for sustained production. *Onion News Tropics*, 4: 43-44.
- Dubois, M., Smith, F.; Gilles, K.A.; Hamilaton, J.K. and Rebers, P.A. 1956. Colorimetric method for determination of sugars and related substances. *Anal. Chem.*, 28(3): 350-356.
- Elad, Y. 2000. Biological control of foliar pathogens by means of *Trichoderma harzianum* and potential modes of action. *Crop Prot.*, **19**:709-714.
- Fayzalla, S.A.; Metwally, A.H. and Sadat, M.M. 2011. Effect of some fungicides, bioagents, essential oils for controlling purple blotch disease of onion. *J. Plant Prot. Pathol.*, Mansura Univ., 2(2): 663-676.
- Gupta, R.B.L. and Pathak, V.N. 1988. Yield losses in onions due to purple leaf blotch disease caused by *Alternaria porri*. *Phytophylactica*, **20**: 21-23.
- Harman, G.E.; Howell, C.R.; Viterbo, A.; Chet, I. and Lorito, M. 2004. *Trichoderma* species-opportunistic, a virulent plant symbionts. *Nat. Rev. Microbiol.*, 2: 43-56.
- Harman, G.E. 2006. Overview of mechanisms and uses of *Trichoderma* spp. *Phytopathology*, **96**: 190–194.
- Hasan, A.; Nisha, H.A.C.; Belal H., M.d. and Rafiqul Islam, M.d. 2016. Evaluation of combined effect of micronutrients (ZnSO₄ + Borax) and fungicides to control the purple blotch complex of onion (*Allium cepa*). *American J. Plant Sci.*, 7: 715-723.
- Jackson, M.L. 1967. Soil Chemical Analysis, Prentice Hall of India, New Delhi, pp144-197.
- Javadzadeh, A.; Ghorbanihagho, A.; Bonyadi, S.; Rashidi, M.R.; Mesgari, M.; Rashtchizadeh, N. and Argani, H. 2009. Preventaion effect of onion juice on selenite-induced experimental cataract. *Ind. J. Ophthalmol.*, 57: 185-189.
- Kumar, P.T. 2007. Biological management of Alternaria blight of onion. M. Sc. College of Agriculture, Dharwad University of Agricultural Sciences, Dharwad, India. 112p.
- Kumar, P.T. and Palakshappa, M.G., 2008. Management of purple blotch of onion through bioagents. *Karnataka J. Agric. Sci.*, **21**(2): 306-308.
- Lakra, B.S. 1999. Development of purple blotch incited by *Alternaria porri* and its losses in seed crop of onion (*Allium cepa*). *Indian J. Agric. Sci.*, **69**(2): 144-146.
- Prakasam, V. and Sharma, P. 2012. *Trichoderma harzianum* (Th-3) a potential strain to manage the purple blotch of onion (*Allium cepa* L.) caused by *Alternaria porri* under North Indian Plains. J. Agric. Sci., 4: 266-272.

- Qadri, S.M.H.; Srivastava, K.S.; Bhope, S.R.; Pandey, U.B. and Bhagchan Dani, P.M. 1982. Fungicidal bioassay against some important pathogens of onion. *Pesticides*, 16: 11-16.
- Rahman, M.L.; Ahmed, H.U. and Mian, I.H. 1988. Efficacy of fungicides in controlling purple blotch of onion. *Pl. Pathol.*, 4: 71-76.
- Rial-Otero, R.; Arias-Estévez, M.; López-Periago, E.; Cancho-Grande, B. and Simal-sGándara, J. 2005. Variation in concentrations of the fungicides tebuconazole and dichlofluanid following successive applications to greenhousegrown lettuces. J. Agr. Food Chem., 53: 4471-4475.
- Riker, A.J. and Riker, R.S. 1936. Introduction To Research On Plant Diseases, St. Louis, Chicago, New York and Indianapolis, John's Swift Co., 117p.
- Schwartz, H.F. 2004. Botrytis downy mildew and purple blotch of onion. Colorado State University Cooperative Extension, **2**: 941.
- Sharma, S.R. 1986. Effect of fungicidal sprays on purple blotch and bulb yield of onion. *Indian Phytopath.*, **39**(1):78–82.
- Singh, R.S. 1982. Plant Pathogens "The Fungi". Oxford and IBH Publishing Co.New Delhi, Bombay, Calcuta, 443p.
- Sliva, G.H.; Costa, V.P.; Campos, V.P.; Oliverira, D.F. and Pfenning, L.H. 2001. Fungal metabolites with activity against nematodes. Bioactive Fungal Metabolites. Impact and Exploitation, International Symposium. Br. Mycolog. Soc., Wales Swansea, UK, 95p.
- Snedecor, G.W. and Cochran, W.G. 1989. Statistical Methods, 8th ed. Iowa State Univ. Press, Ames, Iowa USA, 503p.
- Surviliene, E.; Valiuskaitė, A. and Raudonis, L. 2008. The effect of fungicides on the development of downy mildew of onions. *Zemdirbyste Agric.*, 95(3): 171-179.
- Vijayalakshmi, M.; Madhavi, M. and Kavitha, A. 2012. Studies on Alternaria porri (Ellis) Ciferri pathogenic to Onion (Allium cepa L). Archives Applied Sci. Res., 4(1):1-9.

(Received 23/01/2017; in revised form 27/02/2017)

تأثير بعض المعاملات الحيوية في مقاومة مرض اللطخة الأرجوانية على البصل في مصر محمد فاروق عطية أحمد× ــ محسن محمدى أمين×× ــ إبراهيم عبد المنعم الفقي××× *المعمل المركزي للزراعة العضوية - مركز البحوث الزراعية ** معهد بحوث أمراض النباتات - مركز البحوث الزراعية -مصر. *** كلية الزراعة بمشتهر - جامعة بنها – مصر.

أجريت هذه الدراسة لمدة سنتين متتاليتين خلال موسمي النمو ٢٠١٥/٢٠١٤ و ٢٠١٦/٢٠١٥ تحت الظروف البيئية الطبيعية لنمو نباتات البصل على صنفي بصل جيزة ٢٠ و جيزة أحمر بمزرعة بايو لاند - محافظة الشرقية - مصر. تم تقييم كفاءة بعض المعاملات الحيوية مثل ترايكودرما هايرزيانم، ترايكودرما فيردي، ترايكودرما هاماتم و نوع من كيتوميم كمعاملة رش للمجموع الخضري في وجود المبيد الحيوى بلايت استوب و المبيد الكيماوي الموصى به لونا اكسبرينس كمقارنة لمكافحة مرض اللطعة الأرجوانية. سجلت جميع المعاملات أعلى أفضلية في خفض شدة الإصابة لمرض اللطخة الأرجوانية على نباتات البصل مقارنةً بالنباتات غير المعاملة. كانت المعاملة بالفطر ترايكودرما هايرزيانم أفضل المعاملات و تفوَقت على المبيد الحيوي بلايت استوبٌ و على النقيضٌ تماماً أظهرت المعاملة بِالفطر كيتوميم أقل تأثير. علاوة على ذلك فإن جميع المعاملات السابقة سجلت أعلى زيادة في المحصول، ارتفاع النبات، عدد الأوراق، الوزن الرطب، محتوى الأبصال من الكربوهيدرات الكلَّية، النيتروجين الكلى و نسبة الصلابة مقارنة بالنباتات غير المعاملة